Incorporation of C^{14}-labeled AMP into ATP is the most direct evidence for reversibility of amino acid incorporation into RNA. No incorporation of AMP into ATP occurred when RNA plus free amino acid was used in place of amino acid-RNA, or when P–P was omitted. Calculation of the data shows that the molar rate of AMP incorporation into ATP greatly exceeds the rate of amino acid-RNA breakdown, suggesting that reactions which form unlabeled ATP occur during the incubation.

The approximate equilibrium position of these reactions was near unity, indicating the high energy nature of the amino acid-RNA linkage.

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16-ALKYLATED CORTICOIDS. I. 16α-METHYLPREDNISONE AND 16β-METHYLPREDNISONE[1]

Sir:

The recent publication by Boland[2] on 16α-methyl corticosteroids prompts us to report our studies of both 16α- and 16β-methyl steroids which possess a high order of anti-inflammatory activity without salt retention in animal and clinical trials.

Reaction of 3α-acetoxy-16-pregnene-11,20-dione[3] (I) with diazomethane gave an intermediate pyrazoline,[4] m.p. 199–200° dec., $[\alpha]_D$ +149.6° (all rotations 1% in diox.). *Anal.* Found: C, 69.51; H, 7.98; N, 6.69. Pyrolysis of this product at its melting point gave 3α-acetoxy-16-methyl-16-pregnene-11,20-dione, m.p. 163–166°, $[\alpha]_D$ +69.9°, λ_{max}^{MeOH} 248 mμ (ε 10,800). *Anal.* Found: C, 74.58; H, 8.55. Reduction with palladium yielded 3α-acetoxy-16β-methyl pregnane-11,20-dione (II) m.p. 160–163°, $[\alpha]_D$ +93.6°, no ultraviolet absorption between 220–300 mμ. *Anal.* Found: C, 74.37; H, 9.06. Enol acetylation with *p*-toluenesulfonic acid and acetic anhydride, then treatment with peracetic acid and finally alkaline hydrolysis, gave 3α,17α-dihydroxy-16β-methylpregnane-11,20-dione, m.p. 182–185°, $[\alpha]_D$ +83.6°. *Anal.* Found: C, 72.82; H, 8.92. Bromination at C-21 and then treatment with potassium acetate gave 3α,17α-21-trihydroxy-16β-methylpregnane-11,20-dione 21-acetate, m.p. 200–206°, $[\alpha]_D$ +119.6°. *Anal.* Found: C, 68.79; H, 8.39. Oxidation with N-bromosuccinimide produced the 3-ketone, m.p. 198–202°, $[\alpha]_D$ +128.0°. *Anal.* Found: C, 69.04; H, 8.10. Dibromination at positions 2 and 4, followed by dehydrobromination with dimethylformamide, produced 16β-methylprednisone 21-acetate (III) m.p. 232–235°, $[\alpha]_D$ +213.6°, λ_{max}^{MeOH} 238 mμ (ε 14,200). *Anal.* Found: C, 69.24, H, 7.21. Hydrolysis with potassium bicarbonate gave 16β-methylprednisone, m.p. 210–204°, $[\alpha]_D$ +190.2°, λ_{max}^{MeOH} 238 mμ (ε 14,700). *Anal.* Found: C, 71.19; H, 7.37

Reaction of I with methylmagnesium iodide produced 3α-hydroxy-16α-methylpregnane-11,20-dione,[5] m.p. 149–154°, $[\alpha]_D$ +100.5°, no selective absorption between 200 and 340 mμ. *Anal.* Found: C, 74.16; H, 9.41. This was converted into 16α-methylprednisone 21-acetate (m.p. 212–214° $[\alpha]_D$ +157.8°, λ_{max}^{MeOH} 238 mμ (ε 15,500). *Anal.* Found: C, 69.84; H, 7.22) by the same procedure used for the conversion of II to III (*i.e.*, enol acetylation and peroxidation, 21-bromination and acetoxylation, oxidation at C-3 and 2,4-dibromination and dehydrobromination).

A direct comparison of prednisone and its 16-methyl derivatives in human subjects utilizing (1) metabolic balance studies[6] consisting of the analysis of diet, urine and feces for calcium, phosphorus, nitrogen, sodium and potassium, and (2) the clinical response of patients indicate that 16-methylation (α or β) of the parent steroid, prednisone, is associated with an enhancement of anti-anabolic properties and an increase of 30–50% in both anti-inflammatory and sodium excreting properties. Unlike 16α-hydroxylation, 16α- or 16β-methylation contributes to anti-inflammatory potentiation. The *in vivo* conversion of the 16-methyl corticoids into urinary 17-keto steroids is limited to less than 5%: a conversion slightly less than that obtained with the parent steroids unsubstituted at position 16, and much less than that obtained with cortisone or hydrocortisone.

(1) After submission of this manuscript, a Communication appeared [G. Arth, D. Johnston, J. Fried, W. Spooncer, D. Hoff and L. Sarett, THIS JOURNAL, **80**, 3160 (1958)] describing the preparation of 16α-methylprednisone by essentially the same route. We have tried to eliminate as much of the common material as possible.

(2) E. W. Boland, *Cal. Med.*, **88**, 417 (1958).

(3) H. Slates and N. Wendler, *J. Org. Chem.*, **22**, 498 (1957).

(4) *Cf.* A. Wettstein, *Helv. Chim. Acta*, **27**, 1803 (1944).

(5) *Cf.* R. Marker and H. Crooks, THIS JOURNAL, **64**, 1280 (1942).

(6) E. C. Reifenstein, F. Albright and S. Wells, *J. Clin. Endocrinol.*, **5**, 367 (1945).

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THE INTERMEDIATE COBALT HYDROCARBONYL–OLEFIN COMPLEX IN THE OXO REACTION[1]

Sir:

The several mechanisms[2,3,4] which have been suggested for the oxo synthesis all involve a rate-determining displacement of a mole of carbon monoxide from a carbonyl of cobalt by the attacking olefin. The present study shows that not only does complexing occur between olefin and hydrocarbonyl[5] under room conditions without the libera-

(1) We wish to thank the Houdry Process Corp. for a generous fellowship which made this work possible.

(2) H. W. Sternberg, R. Markby and I. Wender, THIS JOURNAL, **79**, 6116 (1957); I. Wender and M. Orchin, in "Catalysis," Vol. V, Reinhold Publishing Corp., New York, N. Y. 1957, p. 124.

(3) A. R. Martin, *Chem. and Ind.*, 1536 (1954).

(4) G. Natta, R. Ercoli, S. Castellano and P. H. Barbieri, THIS JOURNAL, **76**, 4094 (1954).

(5) M. Orchin, L. Kirch and I. Goldfarb, *ibid.*, **78**, 5450 (1956).